

CLAIMS

[1] A method for screening genomic DNA fragments capable of providing plants with an agriculturally advantageous phenotypic variation, comprising the steps of:

1) preparing genomic DNA from a plant, which is then cloned into a cloning vector to form a genomic DNA library;

2) introducing the genomic fragment from each of the genomic clones constituting the genomic DNA library separately into a plant to produce transgenic plants;

3) cultivating the transgenic plants or progeny thereof to select a plant exhibiting an agriculturally advantageous phenotypic variation;

4) selecting the genomic DNA fragment, which was introduced in step (2) into the plant selected in step (3), as a purposed genomic DNA fragment; and

5) optionally introducing the genomic DNA fragment selected in step (4) or a part thereof into a plant to repeat steps (3) and (4), and selecting a genomic DNA fragment which produces a plant exhibiting an agriculturally advantageous phenotypic variation as a purposed genomic DNA fragment in each repetition.

[2] A method according to claim 1, wherein the size of the selected genomic DNA fragment is 1 kb or greater provided that the DNA fragment can be introduced into the cloning vector.

[3] A method for screening according to claim 1 or 2,

wherein step (2) comprises the sub-steps of: introducing the genomic fragment into the genome of a cell or tissue of the plant; regenerating a complete plant from the plant cell; and cultivating the regenerated plant.

[4] A screening method according to claim 3, wherein the introduction of the genomic DNA fragment into a plant cell or tissue is conducted by a method selected from the group consisting of biological introduction methods, physical introduction methods and chemical introduction methods.

[5] A screening method according to any one of claims 1 to 4, wherein the agriculturally advantageous phenotypic mutation in a plant gives rise to an increase or decrease of the size or the weight of at least a part of the plant or of at least a constituent thereof, an increase of growth rate or an excellent resistance against diseases or pests, under normal cultivation conditions, as compared with a case where the plant does not have the phenotypic variation.

[6] A screening method according to any one of claims 1 to 4, wherein the agriculturally advantageous phenotypic variation in a plant gives rise to an increase or decrease of the size or the weight of at least a part of the plant or of at least a constituent thereof, an increase of growth rate or an excellent resistance against diseases or pests, under conditions which are more stressful for the plant than normal conditions, as compared with a case where the plant does not have the phenotypic variation.

[7] A screening method according to claims 5 or 6,

wherein the plant transformed in step (2) is of the same species as that of the plant which supplied the genomic DNA in step (1).

[8] A screening method according to claim 5 or 6, wherein the plant transformed in step (2) is of a different species from that of the plant which supplied the genomic DNA in step (1).

[9] A screening method according to claim 7 or 8, wherein the optional introduction of the genomic DNA fragment in step (5) is made into a plant of the same species as that of the plant which was transformed in step (2).

[10] A screening method according to claim 7 or 8, wherein the optional introduction of the genomic DNA fragment in step (5) is made into a plant of a different species from that of the plant which was transformed in step (2).

[11] A screening method according to claim 9 or 10, wherein the plant subjected to the optional introduction of the genomic DNA fragment in step (5) is cultivated under the same conditions as in the plant transformed in step (2).

[12] A screening method according to claim 9 or 10, wherein the plant subjected to the optional introduction of the genomic DNA fragment in step (5) is cultivated under different conditions from those in the case of the plant transformed in step (2).

[13] A method for producing a genomic DNA fragment capable of bringing about an agriculturally advantageous phenotypic variation in plants comprising the steps of:

culturing E.coli cells containing a cloning vector carrying a genomic DNA fragment selected by the method according to any one of claims 1 to 12; and preparing the cloning vector amplified in the E. coli cells along with the genomic DNA fragment.

[14] A method for producing a genomic DNA fragment wherein the genomic DNA fragment selected by the method according to any one of claims 1 to 12 is used as a template and the amplification of the fragment is conducted by a biochemical amplification method.

[15] A method for producing a DNA fragment wherein the genomic DNA fragment obtained by the method of claims 13 or 14 is digested with restriction enzyme(s).

[16] A DNA fragment produced by the method according to any one of claims 13 to 15.

[17] A method for producing a plant having an agriculturally advantageous phenotypic variation comprising the step of introducing a genomic DNA fragment capable of bringing about an agriculturally advantageous phenotypic variation in plants, wherein the genomic DNA fragment is produced by a method comprising the steps of: culturing E. coli cells containing a cloning vector carrying the genomic DNA fragment, and preparing the cloning vector amplified in the E. coli cells along with the genomic DNA fragment.

[18] A method for producing a plant having an agriculturally advantageous phenotypic variation according to claim 17 wherein the step of introducing a genomic DNA

fragment capable of bringing about an agriculturally advantageous phenotypic variation in plants comprises the steps of: introducing the genomic fragment into a plant cell or tissue; regenerating a complete plant from the plant cell; and cultivating the regenerated plant.

[19] A method for producing a plant according to claim 18 wherein the introduction of the genomic DNA fragment into a plant cell or tissue is conducted by a method selected from the group consisting of biological introduction methods, physical introduction methods and chemical introduction methods.

[20] A method for producing a plant according to any one of claims 17 to 19, wherein the genomic DNA fragment capable of bringing about an agriculturally advantageous phenotypic variation is introduced in a plant of the same species as that of the plant from which the genomic DNA fragment was derived.

[21] A method for producing a plant according to any one of claims 17 to 19, wherein genomic DNA fragment capable of introducing an agriculturally beneficial phenotypic variation is introduced in a plant of a different species from the plant from which the genomic DNA fragment was derived.

[22] A plant produced by a method according to any one of claims 17 to 21.

[23] A method for analyzing a genomic DNA fragment capable of bringing about an agriculturally advantageous phenotypic variation comprising the steps of: culturing

E. coli cells containing a cloning vector carrying a genomic DNA fragment selected by the method according to any one of claims 1 to 12; and preparing the cloning vectors amplified in the E. coli cells along with the genomic DNA fragment, and reading the nucleotide sequence of the plant genomic DNA fragment in the cloning vector.

[24] A method for analyzing a DNA fragment comprising the step of restricting the genomic DNA fragment selected according to any one of claims 1 to 12.

[25] A method for analyzing a DNA fragment wherein the genomic DNA fragment selected by the method according to any one of claims 1 to 12 is used as a template and the amplification is conducted by a biochemical amplification method.

[26] A method according to claim 24 or 25 wherein the analysis comprises the step of reading the nucleotide sequence of the restriction product of the genomic DNA fragment or the biochemically amplified product.

[27] A method of using a genomic DNA fragment selected by the method according to any one of claims 1 to 12 as a marker to be used in the improvement of a plant variety.

[28] A method according to claim 27 wherein a genomic DNA fragment selected in accordance with any one of claims 1 to 12 is used as a marker, wherein presence of the marker in the genomic DNA from a plant indicates that the plant is useful for improving a plant variety whereas a plant not having the marker is not useful for improving a plant variety.

[29] A method according to claim 28 wherein a genomic DNA preparation is prepared from a progeny plant which is obtained by crossing a plant known to contain a genomic DNA fragment selected according to the method of any one of claims 1 to 12 with a plant of a variety to be improved, and if a progeny plant contains the same genomic DNA fragment, said progeny plant is useful for a further step of improvement of the plant variety.

[30] A method according to claim 29 wherein the plant known to contain a genomic DNA fragment selected according to the method of any one of claims 1 to 12 is a plant as claimed in claim 22.